23 PSILOCYBIN AND PSILOCYN METHODOLOGY	Page 1 of 1
Division of Forensic Science	Amendment Designator:
CONTROLLED STUBSTANCES PROCEDURES MANUAL	Effective Date: 9-December-2003

23 PSILOCYBIN AND PSILOCYN METHODOLOGY

23.1 Scheduling:

- 23.1.1 Schedule I psilocybin and psilocyn, which are found in mushrooms.
- 23.1.2 The chemicals, rather than the botanical, are controlled.

23.2 Extractions:

- 23.2.1 Dry sample (in drying oven or microwave). Grind and soak in methanol for a period of 1-24 hours. Filter off plant material prior to analysis. This extraction method will allow for the analysis of either psilocyn or psilocybin.
- 23.2.2 Acetic Acid Extraction Technique (recommended for mushrooms in chocolate or other matrices). This extraction will allow for the analysis of psilocyn.
 - Dry approximately 3 grams material. (Easier to grind when dry.)
 - Grind up.
 - Let soak in 6% acetic acid for 30 minutes 1 hour.
 - Filter off insoluble material.
 - Extract acid portion with three aliquots of CHCl₃.* (Discard CHCl₃.)
 - Basify acid portion with concentrated NH₄OH to pH 8 10.
 - Extract basic solution with three aliquots of CHCl₃.*
 - Combine aliquots of CHCl₃.
 - Evaporate CHCl₃ with air (low heat).
 - Resultant residue will yield psilocyn.

23.3 Color Test Results:

- 23.3.1 Ehrlich's- purple (positive for psilocyn and psilocybin).
- 23.3.2 Weber Test- psilocyn Fast Blue B or Fast Blue BB gives red color; addition of conc. HCl gives blue color.

23.4 TLC:

- 23.4.1 Baths: TLC1, TLC2, TLC3, TLC4 and TLC5 are recommended.
- 23.4.2 Detection Sprays:
 - 23.4.2.1 p-DMAB and HCl reddish violet area for psilocybin, blue for psilocyn.
 - 23.4.2.2 Weber Test (Fast Blue B or Fast Blue BB) can also be used as a TLC spray to detect psilocyn.

23.5 GC:

- 23.5.1 GC and GC/MS will give only psilocyn due to the dephosphorylation of the psilocybin caused by the GC injection port temperatures.
- 23.5.2 After methanol extraction and drying, silylation with BSTFA prior to GC or GC/MS will allow differentiation of psilocybin from psilocyn. If psilocybin is to be confirmed, derivatization is required.
- **23.6 FTIR:** KBr pellet on extract from acetic acid extraction yields psilocyn.

^{*} Do not mix vigorously in the separatory funnel as an emulsion will probably form.